REMARKS

Summary of the Invention

The invention, as delimited in the present claims, features a method for detecting an increased risk of developing Down's Syndrome, cardiovascular disease, or cancer in a mammal (e.g., an embryo or fetus) by detecting the presence of a G instead of an A at position 66 relative to the first nucleotide of the start codon of MTRR in the mammal or a future parent of the mammal. The invention also features a method for detecting an increased risk of a folate/cobalamin metabolic disorder in a mammal by detecting the presence of a G instead of an A at position 66 relative to the first nucleotide of the start codon of MTRR, a deletion of 4 nucleotides starting from position 1675 (nucleotides 1675-1678) relative to the first nucleotide of the start codon of MTRR, or a deletion of 3 nucleotides starting from nucleotide 1726 (nucleotides 1726-1728) relative to the first nucleotide of the start codon of MTRR. Also featured is a method for detecting an increased risk of developing a neural tube defect in a mammalian embryo or fetus by detecting a G instead of an A at position 66 relative to the first nucleotide of the start codon of MTRR and a low serum cobalamin level in the embryo or fetus or a future parent of the embryo or fetus.

Summary of the Office Action

Claims 6-9, 11-21, and 35-43 are pending. Claims 12 and 15-20 are withdrawn from consideration. Claims 6-9, 11, 13, 14, 21, and 35-43 are rejected under 35 U.S.C. § 112, first paragraph, for lack of enablement. By this reply Applicants cancel claims 40 and 41, amend

claims 6, 35, and 42, add new claims 44-54, and address each of the Examiner's rejections below.

Support for the Amendment

Support for the amendment to claims 6, 35, and 42 is found in prior claims 6, 35, and 42. Support for new claims 45-49 is found in the specification on, e.g., page 4, line 19, through page 5, line 8, page 30, line 5, through page 33, line 16. Support for new claims 44 and 50-54 is found in the specification on, e.g., page 8, line 14, through page 9, line 22, page 12, line 8, through page 13, line 20, and page 28, lines 19-23, and page 56, line 10, through page 58, line 23. No new matter has been added by the amendment.

Rejections under 35 U.S.C. § 112, first paragraph

Claims 6-9, 11, 13, 14, 21, and 35-43 are rejected under 35 U.S.C. § 112, first paragraph, for lack of enablement. The Examiner acknowledges that the specification is enabling for demonstrating the correlation between an increased risk of neural tube defects (NTD) by detecting the presence of the homozygous A66G methionine synthase reductase (MTRR) genotype and a low cobalamin level. The Examiner also acknowledges that the specification demonstrates a correlation between the A66G MTRR mutation and an increased risk for Down's Syndrome and coronary artery disease (CAD). The Examiner states that the specification:

> does not reasonably provide enablement for a method for detecting an increased risk of developing a NTD, Down's Syndrome, hyperhomocysteinemia, cancer or cardiovascular disease in any mammalian fetus or embryo by detecting any heterozygous or homozygous MTRR polymorphism in either or both future

parents of said embryo or fetus, or in said embryo or fetus. Office Action, pp. 2-3. Applicants respectfully disagree, but in the interest of expediting prosecution of the claims, Applicants have amended claims 6 and 35 to recite a method for detecting an increased risk of developing Down's Syndrome, cardiovascular disease, or cancer by detecting the presence of the A66G MTRR polymorphism only. Applicants have also added new claim 44, which recites that the method of claim 6 or 35 detects an increased risk for colon cancer; claims 45-49, which are directed to a method for detecting an increased risk of a folate/cobalamin metabolic disorder by detecting the presence of either an A66G MTRR polymorphism, a deletion of 4 nucleotides starting from position 1675 (nucleotides 1675-1678) relative to the first nucleotide of the start codon of MTRR, or a deletion of 3 nucleotides starting from nucleotide 1726 (nucleotides 1726-1728) relative to the first nucleotide of the start codon of MTRR; and new claims 50-54, which are directed to a method for detecting an increased risk of developing a neural tube defect in a mammalian embryo or fetus by detecting the presence of an A66G MTRR polymorphism and low serum cobalamin. For the reasons discussed below, these amendments overcome the present rejection of claims 6-9, 11, 13, 14, 21, and 35-43 under 35 U.S.C. § 112, first paragraph, for lack of enablement.

Amended claim 6 now recites a method for detecting an increased risk of developing Down's Syndrome, cardiovascular disease, or cancer in a mammalian embryo or fetus by detecting the presence of a homozygous MTRR polymorphism consisting of a G instead of an A at position 66 relative to the first nucleotide of the start codon of MTRR in an embryo or fetus or in a future parent of an embryo or fetus, or by detecting the presence of a heterozygous or

homozygous A66G MTRR polymorphism in both future parents of the embryo or fetus. The Examiner acknowledges that the specification is enabling for a claim to a method for detecting an increased risk of cardiovascular disease in an individual by detecting a homozygous A66G MTRR polymorphism in that individual (see Office Action, p. 3), but states that the specification does not reasonably provide enablement for a method that detects an increased risk for Down's Syndrome or cancer in an individual by detecting a homozygous A66G MTRR polymorphism in that individual. Applicants respectfully disagree.

The Specification is Enabling for Detection of Down's Syndrome by Detecting an A66G MTRR Polymorphism

Applicants direct the Examiner to page 63, line 4, through page 64, line 1, which states:

> We found that mothers of Down's Syndrome babies had a significant 2.49 fold greater likelihood of having a homozygous mutation for the A → G polymorphism at nucleotide position 66. In addition, we found that mothers of Down's Syndrome babies had a 2.07 fold greater likelihood of having a heterozygous mutation or a homozygous mutation in the MTHFR gene. Finally, we identified a positive interaction between the MTRR and MTHFR gene mutations. Table 6 demonstrates that mothers with Down's Syndrome babies had an even greater likelihood of having both the MTRR and MTHFR mutations than either the MTRR or MTHFR mutations alone. Mothers with Down's Syndrome babies had a 3.71 fold greater likelihood of having both a MTRR and a MTHFR mutation than control mothers. This result indicates that the identified mutations are useful as genetic markers for detection of Down's Syndrome in a fetus or embryo. Alternatively, these mutations can be used to assess the risk of a particular mother of having a Down's Syndrome baby.

This passage clearly states that the presence of either an MTRR A66G mutation or an MTHFR C677T mutation indicates a significantly greater risk for Down's Syndrome than in the absence of either mutation, and that these mutations are individually useful as genetic markers for

detection of Down's Syndrome in a fetus or embryo. Although the specification teaches that the risk of having a Down's Syndrome baby is greater when the mother has both mutations, this teaching does not negate or diminish the finding that there is a significantly increased risk for having a Down's Syndrome baby when the mother has only one of the two mutations. Therefore, contrary to the Examiner's assertion, the specification clearly enables a method for detecting an increased risk for Down's Syndrome by detecting the A66G MTRR polymorphism only in an embryo or fetus or a future parent of an embryo or fetus.

The Specification is Enabling for Detection of Cancer by Detecting an A66G MTRR Polymorphism

The specification is also enabling for the detection of an increased risk of developing cancer by detecting the A66G MTRR polymorphism in a subject. The specification states:

> The cloning of human methionine synthase reductase cDNA enables the determination of the enzymatic mechanism involved in the reductive activation of methionine synthase. Furthermore, it is now possible to identify additional mutations in patients with severe deficiency of the enzyme activity, and to determine whether there exist common amino acid polymorphisms which lead to mildly elevated homocysteine levels. Such elevations may be a risk factor in cardiovascular disease, neural tube defects, and cancer. (Specification, p. 32, line 22, through page 33, line 4.

The Examiner argues that "[t]he specification only indicates that the presence of mutations in MTRR gene **are likely** to be associated with altered risk for...cancer. There is no evidence of record that the mutations recited in the claims are correlated to the increased risk of developing those disease[s]" (Office Action pp. 7-8; emphasis in original). Applicants respectfully disagree.

The specification clearly provides an association between MTRR polymorphisms and an

increased risk for cancer. For example, the specification states:

> ...the invention features a method for detecting sequence variants for methionine synthase reductase in a mammal. The method comprises analyzing the nucleic acid of a test subject to determine whether the test subject contains a mutation or polymorphism in a methionine synthase reductase gene. The presence of the mutation or polymorphism is an indication that the animal has an increased or decreased likelihood of developing...cancer. (Specification, p. 8, lines 14-20.)

Furthermore, Applicants' specification clearly identifies a role for a specific MTRR polymorphism, the A66G MTRR polymorphism, in disrupting the folate/cobalamin metabolic pathway. The specification states:

> The fibroblast cell line WG1401 was the first to show the polymorphism, an A to G substitution at bp 66. WG1401 is from patient B.S.S. 17, with megaloblastic anemia, hyperhomocysteinemia, and mild methylmalonic aciduria. (Specification, p. 45, lines 10-13).

Therefore, because the specification teaches a correlation between MTRR polymorphisms associated with elevated homocysteine levels and cancer risk, and because the specification identifies the A66G MTRR polymorphism as being involved in folate/cobalamin metabolic disorders involving elevated homocysteine, the specification is enabling for a method for detecting an increased risk for developing cancer by detecting the A66G polymorphism in a subject.

In support of the enablement of independent claims 6 and 35, Applicants direct the Examiner to two references published after Applicants' discovery, both of which further support Applicants' contention that the specification is enabling for a method of detecting cancer in a subject by detecting the presence of the A66G MTRR polymorphism. The first reference, Matsuo et al. (Asian Pac. J. Cancer Prev. 3:353-359, 2002; a copy of which is provided),

describes a study investigating whether a correlation between methionine and folate related polymorphisms and colorectal cancer exists. Matsuo et al. conclude that the A66G MTRR polymorphism is a risk factor for colorectal cancer.

The second reference, Stolzenberg-Solomon et al. (Cancer Epidemiol. Biomarkers Prev. 12:1222-1226, 2003; a copy of which is provided), confirms a correlation between the presence of the MTRR A66G polymorphism and esophageal squamous cell carcinoma. Therefore, each of the clinical studies confirm a clear correlation between the presence of the A66G MTRR polymorphism and cancer, consistent with Applicants' teachings. Applicants note that these references are not provided to supplement the present specification, but rather, these references demonstrate unequivocally that Applicants' teachings in the present specification are sufficient to teach one skilled in the art how to make and use Applicants' invention with respect to the detection of cancer.

For all of the foregoing reasons, Applicants submit that the specification provides sufficient enabling disclosure for independent claims 6 and 35, as presently amended, and claims dependent therefrom. Therefore, Applicants respectfully request that the rejection of claims 6-9, 11, 13, 14, 21, and 35-43 under 35 U.S.C. § 112, first paragraph, be withdrawn.

The Specification is Enabling for New Claims 45-49

Applicants have also added new claims 45-49, which are directed to a method for detecting an increased risk of a folate/cobalamin metabolic disorder by detecting the presence of either an A66G MTRR polymorphism, a deletion of 4 nucleotides starting from position 1675 (nucleotides 1675-1678) relative to the first nucleotide of the start codon of MTRR, or a deletion of 3 nucleotides starting from nucleotide 1726 (nucleotides 1726-1728) relative to the first nucleotide of the start codon of MTRR. The Examiner states that "[t]he specification only reports the detection of a 4 bp deletion, 1675del4 in WG788 cell line, and a 3 bp deletion, 1726delTTG, in WG1836 cell line but fails to correlate these mutations with increased risk of developing any NTD, Down's Syndrome, hyperhomocysteinemia, cardiovascular disease, or cancer in a mammal" (Office Action, pp. 6-7). Applicants respectfully disagree.

The 4 bp and 3 bp MTRR deletion mutations described in the specification were identified in three patients characterized as being of the *cblE* complementation group of disorders of folate/cobalamin metabolism, who are defective in the reductive activation of methionine synthase (see page 30, lines 17-20, and page 31, line 20, through page 32, line 21, and page 44, line 19, through page 45, line 9, of the specification). The specification states that the WG788 cell line was obtained from the original *cblE* patient, while the WG1146 cell line was obtained from his younger brother. Both of these cell lines contained the 4 bp MTRR deletion polymorphism (see, e.g., page 44, line 19, through page 45, line 9, and page 54, line 3, though page 55, line 7, of the specification). The third cell line described in the specification, WG1836, was obtained from a third patient and was characterized as having the 3 bp MTRR deletion

polymorphism (see, e.g., page 44, line 19, through page 45, line 9, and page 54, line 3, though page 55, line 10, of the specification).

The identification of the 4 bp and 3 bp MTRR deletion polymorphisms, along with the A66G MTRR polymorphism, in the cells of patients diagnosed with a folate/cobalamin metabolic disorder confirmed Applicants' discovery of the MTRR gene and provides the basis for a method of detecting folate/cobalamin metabolic disorders in patients by detecting the presence of these mutations, as is described in the specification (see, e.g., page 30, line 5, through page 33, line 16) and recited in new claims 45-49. Therefore, the specification is enabling for a method of detecting an increased risk of a folate/cobalamin metabolic disorder, as is recited in new claims 45-49, by detecting the presence of either the A66G MTRR polymorphism, or the 4 bp or 3 bp MTRR polymorphisms described above. For this reason, the rejection of claims 6-9, 11, 13, 14, 21, and 35-43 under 35 U.S.C. § 112, first paragraph, for lack of enablement should not be applied to new claims 45-49.

The Specification is Enabling for New Claims 50-54

The specification is also enabling for new claims 50-54, which are directed to a method for detecting an increased risk of developing a neural tube defect in a mammalian embryo or fetus by detecting the presence of both an A66G MTRR polymorphism and low serum cobalamin. The Examiner acknowledges this by stating:

> The specification of the present application discloses increased risk for mothers to develop [children having] neural tube defects (NTD) with combination of **homozygous** mutant MTRR genotype having an A/G polymorphism at base 66, which yields an isoleucine (22I) or a methionine (22M) respectively at amino acid position 22, and **low cobalamin level**... (Office Action, p. 4; emphasis in original.)

Therefore, Applicants respectfully request that the rejection of claims 6-9, 11, 13, 14, 21, and 35-43 under 35 U.S.C. § 112, first paragraph, for lack of enablement not be applied to new claims 50-54.

CONCLUSION

On the basis of the foregoing amendment and remarks, Applicants respectfully submit that pending claims 6-9, 11, 13, 14, 21, 35-39, and 42-54 are in condition for allowance, and a notification to that effect is respectfully solicited.

Enclosed is a petition to extend the period for replying for two months, to and including April 20, 2004, and a check for the required extension fee.

If there are any charges, or any credits, please apply them to Deposit Account No. 03-2095.

Respectfully submitted,

Date: April 20, 2004

Kristina Bieker-Brady, Ph.D., P.C.
Reg. No. 39,109

Clark & Elbing LLP
101 Federal Street
Boston, MA 02110
Telephone: 617-428-0200
Facsimile: 617-428-7045

\\CLARK-W2K1\documents\50004\50004.003004 Reply to 11.20.03 OA.doc